REMARKS

Status of Claims

Claims 1-10, 13-24, and 27-29 stand rejected. Claims 11, 12, 25 and 26 stand objected to. Claims 1, 3-10, 16 and 18-24 have been amended. Claims 1-29 remain pending.

Amendments to the Claims

Claims 1 and 6 were amended to recite "wherein the block of DNA residues comprises at least one mismatch to the nucleic acid molecule." Support for this amendment can be found, for example, on page 8, lines 6-19.

Claims 1, 3-10, 18, and 21-24 were amended to recite a "nucleotide sequence of interest." Support for this amendment can be found, for example, on page 3, line 13.

Claims 4 and 21 were amended to recite a "selectable marker." Support for this amendment can be found, for example, on page 10, line 3.

No new matter has been added by way of these amendments.

The Rejection of the Claims Under 35 U.S.C. §112, First Paragraph, Should Be Withdrawn

Enablement

Claims 1-8, 13-22, and 27-29 were rejected under 35 U.S.C. § 112, first paragraph, for lack of enablement. This rejection is respectfully traversed.

The Examiner continues to maintain that the teachings provided in the instant disclosure are not representative of the ability to predictably introduce nucleotide conversions into the genome of any plant comprising any previously introduced nucleotide sequence of interest. The Examiner sets forth three lines of reasoning to support this position.

First, the Examiner asserts that the experiments contained in the instant application that demonstrated introducing nucleotide conversions into the target AHAS gene resulted in unexpected sequence changes. Applicants respectfully disagree with this conclusion. The instant specification provides data demonstrating two independent target sequences within the endogenous maize AHAS sequence were modified in a site-specific fashion, thereby conferring

resistance to either imidazolinone or sulfonylurea herbicides. Similarly, two independently inserted GFP transgenes were site-specifically modified *in vivo*. In all cases (100%), the modifications resulted in the expected/desired phenotypes. As discussed on page 29, lines 1-4 of the specification, "over 80% of the modifications produced [by the recited methods] were detected at the expected target nucleotide. The remaining 20% were mutated either at the correct target site but with different nucleotides, or at sites upstream but immediately adjacent to the target nucleotide sequence." Moreover, it is noted that of the 20% of the cells mentioned above, *all* produced a desirable modification at or near the target site and resulted in the desired phenotypic change. Accordingly, contrary to the Examiner's conclusion, the specification successfully teaches creating desired alterations designed to inactivate a gene of interest.

Second, the Examiner asserts "the ability to predictably introduce nucleotide conversions into a target gene within an episomal plasmid is not representative of the ability to introduce such conversions into any target gene previously integrated within the genome of the plant." However, the GFP constructs employed in the Examples of the instant invention are not on an episomal plasmid, but rather the GFP constructs have been stably integrated into the genome. In fact, the instant specification provides data demonstrating two GFP constructs having *different* insertions sites in the genome can be successfully targeted by the claimed methods. Further, it is noted that Examiner failed to provide any evidence establishing that an episomal copy verses a stably integrated transgene would have a different level of effectiveness in the methods of the invention. However, as the Examiner had mischaracterized the data in the instant application, the Examiner assertion is moot.

Third, the Examiner asserts that more than routine experimentation is required to practice the invention. Specifically, the Examiner concludes the success of the method is dependent on "higher order structure of the genome at the place of target gene insertion, target gene structure, and repair mechanism of the host plant." Applicants maintain that undue experimentation is not required to practice the claims invention.

As previously made of record, Applicants have successfully identified accessible target sites in two independent PAT/GFP transgenes having distinct and independent chromosomal positions (see, example 2, page 27-28). Similarly, two different positions in the AHAS gene

were also successfully targeted (see, example 2 and table 2 of the specification). In addition, the specification provides specific guidance for target sites in EPSPS, and moreover, provides general strategies for determining appropriate target sites in other genes. See, for example, page 11, lines 22-29 and page 22, lines 3-23 of the specification.

As further evidence, Applicants provide herewith Kochevenko *et al.* (2003) *Plant Physiology 132*:174-184 (Appendix A) and Beetham *et al.* (1999) *Proc. Natl. Acad. Sci. 96*:8874-8778 (Appendix B). The results provided both in the instant application and in Kochevenko *et al.* and Beetham *et al.* are summarized in Table 1 and clearly establish that only routine experimentation is required to practice the claimed invention.

Table 1.

Reference	Plant system	Target Site Modified	Pages of Reference
Beetham *et al.*	dicot	Pro196 of ALS gene	p. 8777, column 2, paragraph 2
Beetham *et al.*	dicot	Codon 6 of GFP transgene integrated into genome	p. 8778, column 1; p8875, column 1,lines 4-5
Kochevonko *et al*	dicot	Try573 of ALS gene	Table 1, p. 179
Kochevonko *et al*	dicot	Pro196 of ALS gene	Table 1, p. 179
Present application	monocot	Ser621 of AHAS	Table 2, page 26
Present application	monocot	Pro165 of AHAS	Table 2, page 26
Present application	monocot	**nt 2987-2990 (end of coding region) of GFP transgene integrated into genome.	Table 2, page 26; page 28, lines 2-3; page 27, lines 25-35
Present application	monocot	**nt 2987-2990 (end of coding region) of GFP transgene integrated into genome.	Table 2, page 26 page 28, lines 2-3; page 27, lines 25-35

** Experiment performed on two independent GFP transgenic lines. Thus, modification of the GFP constructs represents successful targeting at two independent genomic locations.

As illustrated in Table 1, both Kochevenko *et al.* and Beetham *et al.* demonstrate the successful targeting of various positions in a target gene using chimeric oligonucleotides in dicots. As discussed above, the present application demonstrates a success in monocots. Thus, the data summarized in table 1 provides evidence that the use of the chimeric oligonucleotides is successful in both dicots and monocots, and accordingly, contrary to the assertion by the Examiner, the repair mechanism of the host plant does not result in unpredictability.

Moreover, the Examiner's assertion that the "higher order structure of the genome at the place of target gene insertion [and] target gene structure" render the claimed method unpredictable is also not supported by the data set forth in Table 1. In fact, the inaccuracy of this statement is supported by Kochevenko *et al.* which states on page 181, column 2, lines 21-28:

> In our experiments, application of ALF-588 and ALS-1769 chimeraplasts, mediating conversion at the two different sites inside the ALS gene, resulted in comparable frequency of conversion events. Together with the fact that our chimeras had the equal purity and the same level of GC content, *this suggests that the influence of genomic target sequence on the gene repair in tobacco was not detected* (emphasis added).

See, also the abstract of Kochevenko *et al.* that states "similar number of conversion events with two different chimeras indicates the absence of restricting influence of genomic target sequence on gene repair in tobacco." Thus, one of skill in the art concluded, based on the successful targeting of two sites in the genome of a tobacco plant, that genomic target sequence/location does not influence the success of the method. In view of the conclusions in Kochevenko *et al.* and the data summarized in Table 1 which provided evidence of the successful targeting of *7 independent genomic positions in both monocots and dicots*, Applicants have provided clear evidence that, contrary to the assertions by the Examiner, the higher order of the genome and the target sequence does not render the claimed method unpredictable.

The Examiner further asserts the length of the RNA blocks of chimeric oligonucleotides recited in the methods of the invention render the claimed methods unpredictable. Again, the Examiner has failed to provide evidence to support this assertion. See, MPEP 2164.01. Applicants demonstrate above, the methods of the claimed invention are enabled. If the assertion is maintained, the Examiner is respectfully requested to provide evidence to support this position. Further, if the rejection is based on the Examiner's personal knowledge, a declaration under 37 CFR 1.104(d)(2) is respectfully requested.

The Examiner is reminded that the Federal Circuit has repeatedly stated that enablement is not precluded by the necessity for some experimentation, so long as the experimentation needed to practice the invention is not undue. *In re Wands*, 8 USPQ2d 1400 (Fed Cir 1988).

Furthermore, a considerable amount of experimentation is permissible, if it is merely routine, or if the specification provides a reasonable amount of guidance in which the experimentation should proceed. *Id.* As the target sequence is known and the screening methods are adequately described in the specification, the synthesis and screening of chimeric oligonucleotides is nothing more than routine experimentation analogous to the hybridoma screening the Federal Circuit found acceptable in *In re Wands* (858 F.2d 731,8 USPQ 1400(Fed. Cir. 1988)).

In view of the comments and evidence present above, plant genes can be successfully modified at the nucleotide level with a high degree of precision, by using chimeric oligonucleotides. Applicants respectfully request that the rejection of claims 1-8, 13-22, and 27-29 under 35 U.S.C. § 112, first paragraph, be withdrawn.

Written Description

Claims 1-10, 13-24, and 27-29 were rejected under 35 U.S.C. §112, first paragraph, for lack of written description. This rejection is respectfully traversed.

I. The Examiner asserts that the specification and the claims do not indicate what distinguishing attributes are concisely shared by the members of the genus comprising the recited chimeric oligonucleotides. This rejection is respectfully traversed.

Sufficient written description requires simply the knowledge and level of skill in the art to permit one of skill to immediately envision the product claimed from the disclosure. *Purdue Pharm L.P. v. Faulding In.*, 596 USPQ2d 1481, 1483 (Fed. Cir. 2000) ("One skilled in the art must immediately discern the limitations at issue in the claims."). The Examiner's attention is drawn to the "Guidelines for the Examination of Patent Applications Under the 35 U.S.C 112, ¶1, 'Written Description' Requirement," which clearly state that "possession may be shown in many ways." 66 FR 1099, 1105. Applicants may satisfy the written description requirement by "disclosure of any combination of . . . identifying characteristics that distinguish the claimed invention from other materials and would lead one of skill in the art to the conclusion that the applicant was in possession" of the invention. 66 FR 1099, 1106. Applicants provide below

factual evidence that demonstrates possession of the claimed chimeric oligonucleotides recited in the methods of independent claims 1 and 16.

Claims 1 and 16 recite that the chimeric oligonucleotide comprises "at least a first block of RNA residues and a second block of RNA residues, wherein said first and said second blocks of RNA residues are homologous to said nucleic acid molecule and flank a block of DNA residues wherein the block of DNA residues comprises at least one mismatch to the nucleic acid molecule and said chimeric oligonucleotide being capable of recognizing and implementing a nucleotide conversion in said nucleic acid molecule." Ample structure and function is shared among members of the genus to demonstrate possession of the recited chimeric oligonucleotide including: 1) the spatial relationship of the DNA and RNA blocks; 2) the structure of the RNA blocks shared by members of the genus (i.e., homology with the nucleic acid molecule comprising the nucleotide sequence of interest); 3) the common structure of the DNA blocks shared by members of the genus; and, 4) the common function of the recited chimeric oligonucleotides. Applicants describe each of these functional/structural features in more detail below.

First, the chimeric oligonucleotide recited in claims 1 and 16 share a common spatial relationship of the DNA and RNA blocks. Specifically, the claims recite "at least a first block of RNA residues and a second block of RNA residues, wherein said first and said second blocks of RNA residues are homologous to the nucleic acid molecule and flank a block of DNA residues." Accordingly, the position of each of the recited "blocks" in the chimeric oligonucleotide is clear.

Second, the chimeric oligonucleotide recited in claims 1 and 16 share a common structure within the "RNA blocks". As recited in claims 1 and 16, the chimeric oligonucleotide comprises "at least a first block of RNA residues and a second block of RNA residues, wherein said first and said second blocks of RNA residues are homologous to the nucleic acid molecule." As indicated on page 8, lines 7-19 of the specification, the modifying DNA region is flanked by two specific RNA segments that are homologous to the target sequence. The structure of the RNA segments present in the oligonucleotide simply depends on the nucleic acid molecule being targeted and the

location of the desired alteration in the target sequence. As such, the RNA blocks recited in the claims do possess a common structure (*i.e.*, homology to the target sequence) and also a common function (*i.e.*, sufficient homology to the target to allow for the alteration of the target sequence upon introduction into a plant or a plant cell). All that is required is for one of skill in the art to identify the target sequence of interest, determine the desired alteration, and design the "RNA blocks" accordingly. In fact, the specification provides an extensive number of examples of chimeric oligonucleotides that were designed to target various sequences of interest. See, for examples Figures 1-13 and Examples 1 and 2.

Third, claims 1 and 16 further recite that the chimeric oligonucleotide comprises the common structure of "a block of DNA residues". Page 8, lines 7-19 of the specification provides that the DNA residues present in the oligonucleotide comprise a modifying sequence of DNA, which has at least one mismatch to the target sequence. The DNA blocks therefore have a common function as they act as a modification "templates." To expedite prosecution claims 1 and 16 have been amended to explicitly recite that the block of DNA residues comprises "at least one mismatch to the nucleic acid molecule." Moreover, the specification further recognizes that the region of DNA residues employed in the chimeric oligonucleotide will vary depending on the changes that one of skill in the art chooses to introduce into the nucleic acid molecule.

One skilled in the art is clearly apprised of desirable alterations that are capable of inactivating a sequence of interest. For example, in view of the disclosure provided in the specification, chimeric oligonucleotides could be designed to contain a block of DNA residues that introduce a stop mutation or a frame shift mutation into the sequence of interest. Alternatively, the block of DNA residues could be designed to allow for the alteration of regulatory regions that influence expression of the nucleotide sequence of interest. Moreover, it is recognized that one of skill in the art would be familiar with the nucleotide sequence of interest that they are targeting for inactivation and, accordingly, would be aware of additional alterations (i.e., dominant and recessive mutations) that could be made to inactive the sequence. In view of the disclosure in the specification and the general level of knowledge in the art, one of skill in the art would be well apprised of mutations that are capable of inactivating the nucleotide

sequence of interest (i.e., effecting the activity and/or the expression of the sequence in a desired manner) and therefore capable of designing chimeric oligonucleotides comprising an appropriate block of DNA residues that would alter the target sequence accordingly. Thus, the DNA sequences recited in the chimeric oligonucleotide of claims 1 and 16 do share a common structure and function (*i.e.*, have at least one mismatch to a target sequence and produce the desired alteration in a target sequence). In view of the disclosure and general knowledge in the art, one of skill could immediately envision the "DNA block" recited in independent claims 1 and 16 and the respective dependent claims of the instant invention.

Fourth, claims 1 and 16 recite that the chimeric oligonucleotides share a common function. Specifically, claims 1 and 16 further recite that the chimeric oligonucleotide is "capable of recognizing and implementing a nucleotide conversion in said nucleic acid molecule." As previously made of record, routine assays for assessing whether nucleotide conversion has occurred in a target sequence are known in the art. Page 25 of the specification demonstrates an alteration in the target sequence of interest can include an alteration that produces a desired effect on the nucleotide sequence of interest and thereby allows it to be phenotypically selectable. The alteration, however, can further include a change in the target sequence that adds or removes a restriction site. For instance, the example on page 25 of the specification demonstrates *in vivo* mutagenesis using a chimeric ologonucleotide designated AHASG21. The chimeric oligonucleotide was effective at altering both the function of AHAS and destroying a BfaI site. Applicants have therefore demonstrated phenotypic selection and RFLP analysis can be used as an effective screen to assay for the nucleotide conversion. Using a similar approach, Southern blot analysis could also be used to assay for the conversion event. One of skill would further be apprised of assays to screen for gene expression or gene product activity to identify gene inactivation events. Thus, the chimeric oligonucleotides recited in the claimed methods share a common function readily assayed as discussed in the specification.

Accordingly, in view of the common structural and functional characteristics shared by the recited chimeric oligonucleotides, one of skill in the would clearly recognize Applicants were in possession of the chimeric oligonucleotides recited in the claimed methods and thus claims 1-

10, 13-24, and 27-29 satisfy the requirements of 35 U.S.C. §112, first paragraph, and the rejection should be withdrawn.

II. The Examiner further asserts that the specification does not describe the elements that are essential for the definition of "gene". This rejection is respectfully traversed in light of current amendments.

Sufficient written description requires simply the knowledge and level of skill in the art to permit one of skill to immediately envision the product claimed from the disclosure. *Purdue Pharm L.P. v. Faulding In.*, 596 USPQ2d 1481, 1483 (Fed. Cir. 2000) ("One skilled in the art must immediately discern the limitations at issue in the claims."). While Applicants maintain that the term "gene" would be clearly understood by one of skill in the art, to expedite prosecution the claims have been amended to remove the term "gene" and now recite a "nucleotide sequence of interest." The Examiner is respectfully requested to withdraw the rejection of the claims 1-10, 13-24, and 27-29 under 35 U.S.C. §112, first paragraph.

III. The Examiner further asserts that the disclosure fails to describe the characteristics (e.g. nucleotide sequences) to "any and/or all promoters operably linked to any and/or all genes, including any/or all herbicide resistant genes" to allow for the construction of the chimeric oligonucleotide. This rejection is respectfully traversed.

The Examiner is applying an improper standard for written description. "A specification may, within the meaning of 35 U.S.C. §112 ¶ 1, contain a written description of a broadly claimed invention without describing all species that claim encompasses." *Utter v. Hiraga*, 6 USPQ2d 1709 (Fed. Cir. 1988). Accordingly, the Examiner's assertion that the written description requirement has not been satisfied since not all of the species encompassed by the claimed methods have been disclosed is improper.

The assertion that one of skill in the art, in view of the disclosure in the specification, would be unable to identify a target sequence of interest and further be unable to design a chimeric oligonucleotide capable of introducing an alteration that inactivates the sequence of interest has been unsubstantiated by the Examiner. As discussed in detail above, one skilled in

the art is clearly capable of identifying a "sequence of interest" and further is apprised of desirable alterations that are capable of inactivating the sequence of interest, including, for example, stop mutations, frame shift mutations, alterations in regulatory regions that influence expression of the nucleotide sequence of interest, and/or various dominate and recessive mutations.

Moreover, Applicants further note that evidence has been provided above (see arguments related to enablement) that demonstrates the asserted "unpredictability" of the claimed methods is inaccurate. In addition, the Examiner continues to discount the numerous working examples, the prophetic examples, and the guidance provided in the specification for the design of the chimeric oligonucleotides. If the Examiner continues to assert one of skill would not be able to envision a target nucleotide sequence to be used in the instant invention, a further explanation and evidence to support the position is respectfully requested.

In view of the comments above, Applicants submit claims 1-10, 13-24, and 27-29 satisfy the requirement of 35 U.S.C. §112, first paragraph, and the Examiner is respectfully requested to withdraw the rejection.

The Rejection of the Claims Under the Judicially Created Doctrine of Obviousness-Type Double Patenting Should Be Withdrawn.

Claims 1-29 were rejected under the judicially created doctrine of obviousness-type double patenting in view of claims 1, 2, and 4-24 of copending U.S. Application No. 09/580,747. This rejection is respectfully traversed.

The instant application and co-pending U.S. Application No. 09/580,747 are both divisional applications of U.S. Application No. 09/193,612, now U.S. Patent No. 6,528,700. The restriction requirement in U.S. Patent No. 6,528,700 (mailed March 23, 2000) restricted original claims 22-29 (drawn to a method to inactive a gene introduced into a plant genome) and original claims 53-62 (drawn to a method of creating a predetermined nucleotide mismatch in a target sequence in a genome of plant) into two separate Groups. Accordingly, the instant application and Application No. 09/580,747 were filed subject to the restriction requirement of parent case.

The Examiner is reminded that 35 U.S.C. §121 prohibits a double patenting rejection on an application that was filed in response to a Restriction Requirement. See MPEP 804.01. Accordingly, the double patenting rejection is improper and the Examiner is respectfully requested to withdraw the rejection.

The Rejection of the Claims Under 35 U.S.C. §112, Second Paragraph, Should Be Withdrawn

Claims 1-10, 13-24, and 27-29 were rejected under 35 U.S.C. §112, second paragraph, as being indefinite. This rejection is respectfully traversed.

The Examiner states it is unclear how in claims 1 and 16 the blocks of RNA residues can be homologous to the nucleic acid molecule. As explained on page 8, lines 30-15 of the specification "the genetic site of the alteration is determined by selecting a portion of the chimeric oligonucleotide *to have the same sequence as the sequence of the target*. That is, the portion of the chimeric oligonucleotide is *homologous with the target site*. The chimeric oligonucleotides are designed to have at least two homologous regions flanking an interposed heterologous region" (emphasis added). See also, for example, page 3, lines 27-32 which discuss the "homologous paring" between the chimeric oligonucleotide and the plant's target sequence.

Upon review of the disclosure in the specification and in the Figures, one of skill in the art would clearly understand that the homologous structure of the RNA blocks would have a nucleotide sequence corresponding to that of the target sequence with the obvious exception that the RNA blocks would comprise ribonucleic acids and/or variants/derivatives thereof and the homologous target sequence would comprise deoxyribonucleic acids and/or variants/derivatives thereof. It is well established "if the claims, read in light of the specification, reasonably apprise those of skill in the art of both the utilization and scope of the invention, and if the language is a precise as the subject matter permits, the courts can demand no more" *North Am. Vaccine Inc. v. American Cyanamid Co.*, 28 USPQ 2d 1333, 1339 (Fed. Cir. 1993). Claim 6 satisfies this requirement. The Examiner is respectfully requested to withdraw the rejection of claims 1 and 16 under 35 U.S.C. §112, second paragraph.

Claim 16 was rejected for containing a misspelled word. Claim 16 has been amended to have the proper spelling of "oligonucleotide" and the Examiner is respectfully requested to withdraw the rejection.

Consideration Of Previously Submitted Information Disclosure Statement

It is noted that an initialed copy of the PTO Form 1449 that was submitted with Applicants' Information Disclosure Statement filed March 10, 2003 has not been returned to Applicants' representative with the Office Action. Accordingly, it is requested that an initialed copy of the Form 1449 be forwarded to the undersigned with the next communication from the PTO. In order to facilitate review of the references by the Examiner, a copy of the Information Disclosure Statement and the Form 1449 are attached hereto. Copies of the cited references were provided at the time of filling the original Information Disclosure Statement, and, therefore, no additional copies of the references are submitted herewith. Applicants will be pleased to provide additional copies of the references upon the Examiner's request if it proves difficult to locate the original references.

CONCLUSIONS

The Examiner is respectfully requested to withdraw the rejections and allow claims 1-29. Early notice to this effect is solicited.

It is not believed that extensions of time or fees for net addition of claims are required, beyond those that may otherwise be provided for in documents accompanying this paper. However, in the event that additional extensions of time are necessary to allow consideration of this paper, such extensions are hereby petitioned under 37 CFR § 1.136(a), and any fee required therefore (including fees for net addition of claims) is hereby authorized to be charged to Deposit Account No. 16-0605.

Respectfully submitted,

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